

CLAIMS

1. A method for the purification of plasmid DNA in an aqueous two-phase system, comprising the steps of
  - (a) providing a composition comprising a first polymer that exhibits inverse solubility characteristics, a second polymer that is immiscible in the first polymer and optionally a salt;
  - (b) contacting said solution with an aqueous solution comprising plasmid DNA;
  - (c) providing a phase separation and subsequently isolating the aqueous phase;
  - (d) increasing the temperature of the isolated phase to a temperature above the cloud point of the first polymer and below the temperature where plasmid DNA is degraded and subsequently isolating the aqueous phase so formed; and optionally
  - (e) a chromatography step to recover the plasmid DNA from the isolated top phase.
2. A method according to claim 1, wherein the first polymer has a cloud point below about 60°C in the aqueous solution.
3. A method according to claim 1 or 2, wherein the first polymer is selected from the group that consists of polyalkylene glycols, poly(oxyalkylene)polymers, poly(oxyalkylene)copolymers, polyvinyl pyrrolidone, polyvinyl alcohol, polyvinyl caprolactam, polyvinyl methylether, alkoxyLATED surfactants, alkoxyLATED starches, alkoxyLATED cellulose, alkyl hydroxyalkyl cellulose, silicone-modified polyethers, and poly N-isopropylacrylamide and copolymers thereof.
4. A method according to any one of the preceding claims, wherein the first polymer is a copolymer comprised of ethylene oxide and propylene oxide, preferably about 50% of ethylene oxide and about 50% of propylene oxide.
5. A method according to any one of the preceding claims, wherein the second polymer is selected from the group that consists of hydroxyalkyl cellulose, hydroxyalkyl starches, starch, dextran, and pullulan.
6. A method according to any one of the preceding claims, wherein the weight ratio of the amounts of first polymer:second polymer is about 1:1.
7. A method according to claim 6, wherein the amount of the first polymer is about 4.5% (w/w) and the amount of the second polymer is about 4.5% (w/w) of the composition provided in step (a).

8. A method according to any one of the preceding claims, wherein the aqueous solution that comprises plasmid DNA is a cell lysate, which method comprises a step for desalting the cell lysate before step (b).
9. A method according to any one of the preceding claims, wherein the contacting according to step (b) involves mixing at room temperature.
10. A method according to any one of the preceding claims, wherein the isolation according to step (c) and/or step (d) is by centrifugation.
11. A composition for extraction of plasmid DNA in an aqueous two-phase system, which composition comprises a first polymer that exhibits inverse solubility characteristics at temperatures below about 60°C, a second polymer that is immiscible in the first polymer and optionally a salt.
12. A composition according to claim 11, wherein the amount of the first polymer is 4.5% (w/w) and the amount of the second polymer is 4.5% (w/w).
13. A composition according to claim 11 or 12 for separation of plasmid DNA according to any one of claims 1-10.
14. A kit for purification of plasmid DNA from a cell lysate in an aqueous two-phase system, which kit comprises a first polymer that exhibits inverse solubility characteristics at temperatures below about 60°C, a second polymer that is immiscible in the first polymer and optionally a salt in one compartment as well as written instructions for the use thereof.
15. A kit according to claim 14, wherein the first polymer is comprised of ethylene oxide and propylene oxide.
16. A kit according to claim 14 or 15, wherein the second polymer is selected from the group that consists of hydroxyalkyl cellulose, hydroxyalkyl starches, starch, dextran, and pullulan.
17. A kit according to any one of claims 14-16, wherein the weight ratio of the amounts of first polymer:second polymer is about 1:1.
18. A kit according to any one of claims 14-17, which is for purification of a cell lysate that has been desalted before being mixed with an aqueous solution that comprises plasmid DNA.
19. A kit according to any one of claims 14-18 for use in a method according to any one of claims 1-10.

20. Use of a polymer that exhibits inverse solubility characteristics at temperatures below about 60<sup>0</sup>C in an aqueous two-phase system for the purification of plasmid DNA from a cell lysate.

21. Use according to claim 20, wherein the polymer is a copolymer of ethylene oxide and propylene oxide.